

Vermicompost and biochar as substitutes of growing media in ornamental-plant production

J.M. Alvarez^{1,2,3*}, C. Pasian², R. Lal³, R. López⁴ and M. Fernández¹

¹ Escuela Técnica Superior de Ingeniería, Campus La Rábida, Universidad de Huelva, 21071, Huelva, Spain. ² Horticulture Dept., 202 Kottman Hall, Ohio State University, Columbus OH, 43210, USA. ³ C-MASC, 422B Kottman Hall, Ohio State University, Columbus OH, 43210, USA. ⁴ IRNAS-CSIC, Avenida Reina Mercedes, 10, 41012, Sevilla, Spain.

*E-mail: josemaria.alvarez254@alu.uhu.es

Abstract

Vermicompost is a product derived from the accelerated biological degradation of organic wastes by earthworms and microorganisms. Biochar is a by-product of the C-negative pyrolysis technology for bio-energy production from organic materials. Containerized plant production in floriculture primarily utilizes substrates such as peat moss. Environmental concerns about draining peat bogs have enhanced interests in research on complementary products that can be added to peat. Thus, a comparative greenhouse study was conducted to assess the suitability of biochar (*B*) and vermicompost (*V*) as partial substitutes for peat-based growing media for ornamental plant production. Different blends of *B* at a volume fraction of 0, 4, 8, 12 % and *V* at 0, 10, 20, 30, 40, 50 % were compared to a baseline peat substrate (*S*) as control in the cultivation of geranium (*Pelargonium peltatum*) and petunia (*Petunia hybrida*). Substrates were characterized for physical and chemical properties, plant growth, and flower production. Mixtures with low-medium *V* levels (10 -30%) and high *B* level (8 – 12 %) in *Petunia* and *Pelargonium* induced more growth and flower production than that of the control. These results obtained with different *B* and *V* associations are of interest to those who want to reduce peat consumption for the production of ornamental plants in containers and to reduce carbon footprint of this commercially productive sector.

Key words: compost, substrate additive, growing media replacement, carbon storage, *Petunia hybrida*, *Pelargonium peltatum*

Introduction

U.S. 2013 Geological Survey mentioned that worldwide ~11 million metric tons (11 Tg) of *Sphagnum* peat is used annually for horticultural purposes (DOI-USGS, 2013). *Sphagnum* peat moss is the main substrate component used worldwide, because of its consistent physical characteristics (e.g. slow degradation rate, high water holding capacity, and low bulk density) and high nutrient exchange capacity. However, there are increasing environmental concerns because of peat bogs numerous ecosystem services such as valuable habitats, large carbon (C) stocks, water quality and water regime or flood protection (Alexander *et al.*, 2008) which are being affected. Michel in 2010 stated that nowadays there are no satisfactory alternatives to peat, in terms of quality and availability. Nevertheless, some complementary products can be added to peat, especially to improve aeration of the growing medium and enhance nutrient supply. For this reason, numerous studies have been undertaken to assess the potential substitution of peat based substrates with commercial compost and vermicompost, using a range of substitution around 10 to 50 % in volume, for enhancing plant's rooting and growth without any severe negative effects (López *et al.*, 2008). Biochar is a by-product of the C-negative pyrolysis technology for production of bio-energy from organic materials (Lehmann, 2007). Biochar thus produced is not burned for energy production, but is used as a soil amendment to increase water and nutrient retention, lower bulk density, and increase pH (Laird, 2008). Research on biochar has used materials from diverse feedstock and applied to a range of mineral soils for numerous crops and farming systems. Understandably, results available

in the literature are highly diverse and debatable (Jeffery *et al.*, 2011; Lal, 2011; Perry, 2011; Mukherje and Lal, 2014; Lal, 2016). Anyway, some successful experiments have been conducted with ornamental plants in which peat was replaced by biochar (Sohi *et al.*, 2013; Gu *et al.*, 2013; Dispenza *et al.*, 2016). The inclusion of biochar into the substrate created a beneficial environment for microbes, reduced nutrients and water loss and decreased bulk density. Best results have generally been obtained when the recommended dosage of biochar was not greater than 10 to 15 % in volume (Graber *et al.*, 2010; Beck *et al.*, 2011; Tian *et al.*, 2012). Several studies have also reported reduction in the emission of greenhouse gases (GHG) when biochar was used as peat substitute for growing plants (Steiner and Harttung, 2014). Biochar decomposes slowly (Kuzakov *et al.*, 2009) and can be stored for relatively long periods. Above all, data from some experiments also demonstrate the synergistic effects when biochar is combined with compost in the growth medium (Schmidt *et al.*, 2014). Nonetheless, the research information about the effects of biochar blended with compost or vermicompost on substrates devoted to floriculture is scanty and not available. Therefore, the principal objective of this study is to analyze the effects of the vermicompost and biochar added in different ratios and compare to a commercially available peat-based substrate used for the production of petunia (*Petunia hybrida*) and geranium (*Pelargonium peltatum*), and how those ornamental plants will react in growth and flower production. The experiment is designed to test three hypothesis: a) vermicompost and biochar are good component partners to grow petunia and geranium in containers; b) it is possible to define a range of vermicompost

and biochar proportions to produce commercial petunia and geranium plants; c) it is possible to maintain and/or improve the commercial production of these species while reducing the use of substrates from non-renewable sources. We have also considered in this work that it is possible to estimate how much C may be stored for long periods of time when growing *Petunia* and *Pelargonium* in a substrate where growing media has been substituted by vermicompost and biochar.

Material and methods

Organic substrates, plant material and experimental design: One type of vermicompost (*V*) and one type of biochar (*B*) were assayed in this study. The biochar was a commercial product called Soil Reef Pure 02 (Biochar Solutions Inc.) and produced by pyrolysis of *Pinus monticola* wood at high temperature (600 to 800 °C). The vermicompost was also a commercial product from the Black Diamond Vermicompost, and prepared by vermicomposting of dairy manure solids for 70 to 80 days which had been pre-composted for two weeks in an aerated composting system (Table 1, and Tables A.1 to A.3 in the appendix). Both materials were used as organic components to partially replace the normally used standard growing media by the Horticulture Department at the Ohio State University called Farfard 3B mixture by SunGro Horticulture Distribution Inc. (Tables 1, and A.4 in the appendix). Such substrate is composed from the following ingredients: Canadian *Sphagnum* peat moss, processed pine bark, perlite, vermiculite, dolomitic limestone, and a wetting agent, being Peat:Bark:Perlite:Vermiculite volume ratio 6:4:2:1.

Two ornamental species were used in the experiments: *Petunia x hybrida* cv. Dreams Neon and *Pelargonium peltatum* cv. Summer Showers. The choice of cultivars was made based on their responses to substrate electrical conductivity (EC): tolerant for petunias (Mionk and Wiebe, 1961) and sensitive for geranium (Do and Scherer, 2013). Flower production of these two species of ornamental plants was studied because of their major commercial importance.

Treatments consisted of different mixtures of *V* and *B* with the commercially-available peat-based growing mix. Peat-based substrate in the tested mixes received a slow release fertilizer (Scotts Osmocote Plus 15-9-12 at 5.9 g L⁻¹). Twenty four treatments were prepared with the volume fractions detailed in Table 2. Taking into account this design, the separate effects of *V* and *B* could be also deduced by comparing separately the treatments containing *B* = 0 % on the one hand, and *V* = 0 % on the other hand, respectively.

Environment in the greenhouse: The experiment was conducted in the greenhouses of the Department of Horticulture and Crop Science at The Ohio State University, Columbus, OH. *Petunia* and *Pelargonium* plants were first produced in 200 plug trays (21.8 cm³ per plug) for seed germination using a standard germination mix. Two *Petunia* and *Pelargonium* seeds per cell were sown in early February. After germination, just one seedling was kept. Trays were first placed in a germination glasshouse for 43 days at 23.7 °C and 54 % humidity. Seedlings were then transplanted into 15.4 cm diameter plastic containers and moved to a glasshouse (average temperature 20.1 °C and

Table 1. Biochar (*B*), vermicompost (*V*) and peat-based substrate (*S*) characterization. More details of properties of substrate components are shown in the appendix. (Results expressed in dry weight basis except other stated).

Parameter		Biochar	Vermicompost	Peat-based substrate
Organic Matter	(%)	91.6	72.7	55.3
Organic Carbon	(%)	75.8	35.0	n.a.
Total Nitrogen (N)	(%)	0.45	2.90	n.a.
Ammonia (NH ₄ -N)	(mg kg ⁻¹)	5.7	17.0	23
Nitrate (NO ₃ -N)	(mg kg ⁻¹)	64	3100	27
Sulfur (S)	(mg kg ⁻¹)	940	520	18
Sodium (Na)	(%)	0.520	0.300	0.002
Total Potassium (K)	(%)	20.0	0.54	0.01
Total Phosphorus (P)	(%)	0.370	0.436	0.001
EC (1:6 v/v fraction)	(mS m ⁻¹)	37.5	175	14.2
Ph		9.5	6.5	5.47
Total Ash	(%)	8.4	27.3	44.7
Bulk density	(kg dm ⁻³)	0.207	0.131	0.135

n.a.: not analysed.

Plastic containers (15.4 cm diameter, 800 cm³), were filled with each of the mixtures and distributed in a random 5 blocks design for each of the two plant species (2 species x 24 treatments x 5 blocks = 240 containers).

Table 2. Notation used for the substrate mixtures (% in volume of each component): *S*, commercial peat-based growing media; *V*, vermicompost; and *B*, biochar.

Notation <i>S:V:B</i>	Biochar (%)			
	0	4	8	12
Vermicompost (%)				
0	100:00:00	96:00:04	92:00:08	88:00:12
10	90:10:00	86:10:04	82:10:08	78:10:12
20	80:20:00	76:20:04	72:20:08	68:20:12
30	70:30:00	66:30:04	62:30:08	58:30:12
40	60:40:00	56:40:04	52:40:08	48:40:12
50	50:50:00	46:50:04	42:50:08	38:50:12

average humidity 29.3 %) during 8 weeks for *Petunia* and 11 weeks for *Pelargonium*. Standard propagation protocols for these species were followed. Plants were on benches, inside the greenhouse, and occupied 15 m² of surface. Within each block, plants were rotated periodically to minimize variation in microclimatic conditions. Seedlings in plug trays received irrigation by means of a micro sprinkler system and plants in container were watered manually as needed, based on environmental conditions and plant's size under commercial usual conditions, moisture content was kept to field capacity. The entire growing period lasted for 124 days for *Pelargonium* and 90 days for *Petunia*.

Physical and chemical characterization of the substrates: Bulk density (*Db*), container capacity (*Va*), total porosity (*Pt*) and air space (*As*) were determined at the beginning of the experiment following the procedures for determining physical properties of horticultural substrates using the NCSU porometer (Fonteno and Bilderback, 1993). Organic matter was determined by dry ashing at 500 °C. Fresh growing mix samples were used for the determination of soluble nutrients. EC and pH were determined using a 1 to 6 volume fraction aqueous extract (Ansorena Miner, 1994). pH was measured before filtration using a Accumet[®] Ap85 pH-meter. The filtrate was used for EC and mineral-content determinations after extract filtration. EC was determined with a conductimeter (Accumet[®] Ap85). Nitrate-N

and ammonium-N contents were determined in the sample extracts by spectrophotometry in a flow autoanalyser (AA III, Bran + Luebbe, Norderstedt, Germany) (Ansorena Miner, 1994). Total element contents were determined in substrate components by ICP-OES after aqua regia digestion, and were expressed as total contents on a dry matter basis. In substrates, water soluble nutrients were determined by ICP-OES after extraction, and were expressed on a volume basis (Dahlquist and Knoll, 1978). Table A.5 shows pH, EC and mineral nutrients contents of the different substrate mixtures at the beginning of the experiment.

Plant growth and flowering: At the end of the growth period, shoot dry weight (SDW) and number of flowers were recorded. In *Pelargonium* plants, the number of open inflorescences and inflorescence-buds were also counted. Shoot dry weight was obtained after oven-drying at 55 °C for 72 h. Chlorosis and spots in leaves were evaluated using a visual scale ranging from 1 to 10, being 1 a green plant with no chlorosis and no spots, and 10 a yellowish plant or a plant with more than 80 % surface covered by spots (Table A.6).

Leaf nutrient concentration: Plant samples were ground to pass through a 0.5 mm sieve, and then digested by wet oxidation with high purity concentrated HNO₃ under pressure in a microwave oven (Miller, 1998). Mineral nutrients, phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulfur (S), and trace elements iron (Fe), manganese (Mn), boron (B), copper (Cu), zinc (Zn) and sodium (Na), were determined by ICP-OES and expressed on a dry mass basis (Dahlquist and Knoll, 1978). Total nitrogen concentration was determined by spectrophotometry in a flow autoanalyser after Kjeldahl digestion. Plant samples for quality control (WEPAL programs, Houba *et al.*, 1996) were also analyzed. Results obtained for these samples agreed \pm 5 % with the certified results. Tables A7, A8.

Data analysis: One-way analysis of variance (SPSS Statistics 17.0) was carried out to determine statistically significant differences between treatments, being the treatment a fixed effect. Significant differences were established at $\alpha = 0.05$. To compare treatments, Duncan or T3-Dunnnett tests were used in order to differentiate within homogeneous groups (according to variance homoscedasticity), and the Dunnnett test was also used to compare each treatment with the control. In addition, a correlation and regression analysis were performed to establish the underlying relationships between treatments and measured parameters. A two-way ANOVA, with the main effects *V* and *B* and their interaction (*V* \times *B*), was not carried out because *S* content greatly varied by varying *V* or *B*. Likewise, relevant tests of normality and homogeneity of variances were made before proceeding ANOVA, as well as transformation of the data if necessary.

Results

Physical and chemical characteristics of the substrates: The physical properties and OM of *Sphagnum* peat-based substrate (control) *S*, and the different mixtures with biochar (*B*) and vermicompost (*V*) studied are shown in Table 3. Although there are no universally accepted standards for

the physical properties of container substrates, suggested guide ranges are outlined (Fonteno and Bilderback, 1993; Yeager *et al.*, 2000). *Db* and *Va* were always slightly above the recommended range, except for *Db* in the control treatment. *As* in some mixtures (76:20:04, 56:40:04, 72:20:08, 52:40:08, 48:40:12, 38:50:12) was slightly below the optimum range (6-13 %), and were not significantly different from each other.

Table 3. Selected physical properties and OM values of different substrate mixtures (treatments)

Treatment	<i>Db</i>	<i>Va</i>	<i>Pt</i>	<i>As</i>	<i>OM</i>
<i>S:V:B</i> ^x	(kg m ⁻³)	(%)	(%)	(%)	(%)
100:00:00	135 a	70.1 a	81.0 abcde	10.0 e	55.3 a
96:00:04	137 ab	70.8 bc	80.5 abcd	9.8 de	60.7 b
92:00:08	147 bcdef	72.1 abcd	79.8 abcd	7.7 abcde	62.7 bc
88:00:12	146 bcde	72.4 abcde	80.0 abcd	7.6 abcde	66.3 bcde
90:10:00	141 abc	71.0 bc	81.1 abcde	10.1 e	60.6 b
86:10:04	144 abcd	71.0 abc	80.3 abcd	8.8 cde	64.0 bcd
82:10:08	159 ghijk	72.8 bcde	79.0 ab	6.0 abc	65.3 bcde
78:10:12	144 abcd	72.4 abcde	80.0 abcd	7.6 abcde	67.0 cdefg
80:20:00	149 cdefg	75.1 efgh	82.2 cde	7.2 abcde	69.3 efghi
76:20:04	150 cdefg	74.6 defgh	80.3 abcd	5.7 abc	69.3 efghi
72:20:08	163 ghik	73.2 bcdef	78.0 a	5.2 abcd	65.3 bcde
68:20:12	153 defghi	72.3 abcde	80.6 abcd	8.2 bcde	68.3 defgh
70:30:00	154 defghi	74.0 cdefg	81.2 abcde	7.2 abcde	67.0 cdefg
66:30:04	153 defgh	76.4 gh	83.9 e	7.5 abcde	69.3 defgh
62:30:08	156 efghij	73.5 bcdef	79.9 abcd	6.3 abcde	66.7 cdefg
58:30:12	164 ghik	74.8 defgh	81.9 bcde	7.0 abcde	66.7 cdefg
60:40:00	153 defghi	74.7 defgh	82.0 cde	7.3 abcde	69.0 defghi
56:40:04	158 ghijk	74.3 cdefg	79.3 abc	5.1 abc	70.0 efghi
52:40:08	164 hik	75.8 fgh	80.0 abcd	4.2 a	71.3 fghi
48:40:12	180 l	74.9 defgh	79.2 abc	4.4 ab	69.7 defgh
50:50:00	162 hijk	75.8 fgh	82.1 cde	6.2 abcde	71.7 ghi
46:50:04	155 efghij	73.4 bcdef	80.5 abcd	7.0 abcde	72.3 hi
42:50:08	164 ik	77.0 h	83.9 e	6.9 abcde	70.0 efghi
38:50:12	168 k	77.0 h	82.4 de	5.5 abc	73.7 i
<i>p</i>	***	***	***	***	***
Guide ranges ^y	100-300	45-65	78-88	6-13	

Db = Bulk density; *Va* = Container capacity; *Pt* = total porosity; *As* = air space; *OM* = Organic matter.

^x *S:V:B*, Volume fraction of peat based substrate (*S*), vermicompost (*V*) and biochar (*B*). Control treatment = 100:00:00

^y Guide ranges (Landis, 1990; Fonteno and Bilderback, 1993; Yeager *et al.*, 2000; Harp, 2011).

p, significance level: *** indicates $p \leq 0.001$. Different letters in numerical columns indicate significant differences between treatments (Duncan test).

The general trend was a slight but significant decrease in *As* as *V* dose increased in the mixture ($p = 0.012$). Concentration of *V* was inversely related to *As* ($r = -0.43$, $P < 0.01$, $n = 72$), but positively to *Db* ($r = 0.70$, $P < 0.01$, $n = 72$) (Fig. 1) and *Va* ($r = 0.70$, $P < 0.01$, $n = 72$). Nevertheless, there was not significant relationship between *B* and these physical parameters *Db* ($r = -0.15$, $P < 0.01$, $n = 72$); *Va* ($r = 0.11$, $P < 0.01$, $n = 72$); *Pt* ($r = -0.18$, $P < 0.01$, $n = 72$). Treatments with $V \leq 10$ % and $B \leq 4$ % showed no significant differences in *Db* with the control treatment. The latter differed significantly ($P = 0.003$) from all other treatments containing $V \geq 20$ % regardless of the amount of *B* in the mixture. *Pt* of the 24 treatments lay within guide ranges, and the control treatment did not differ significantly in *Pt* from other treatments.

pH was slightly acidic (5.47) for commercial peat-based substrate and gradually increased (up to 6.57 at mixture 38:50:12) as vermicompost

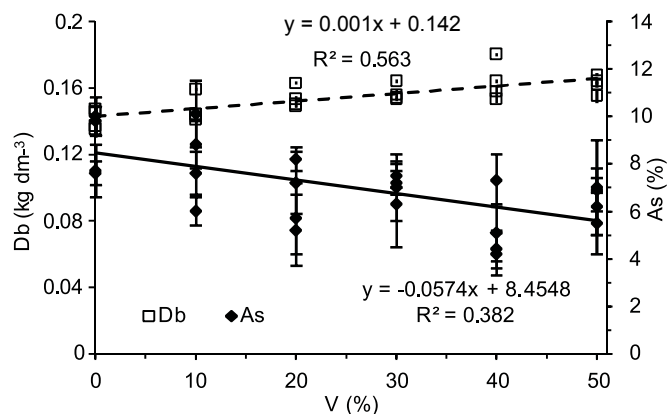


Fig. 1. Relationships between vermicompost (V) content in the substrate and its bulk density (Db) and air space (As). For Db and As mean values (\pm SE) are shown ($n = 24$).

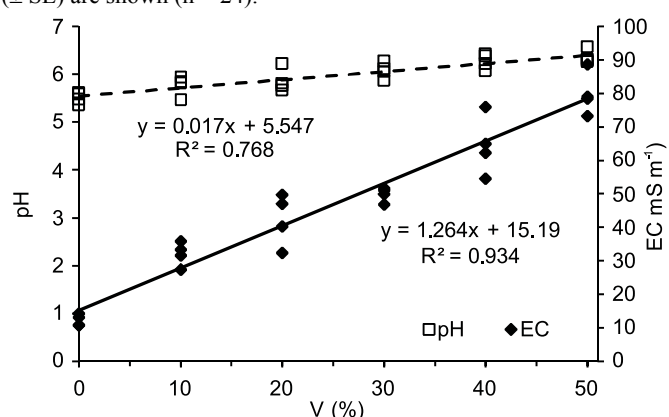


Fig. 2. Relationships between vermicompost (V) content in the substrate and its pH and electrical conductivity (EC), ($n = 24$).

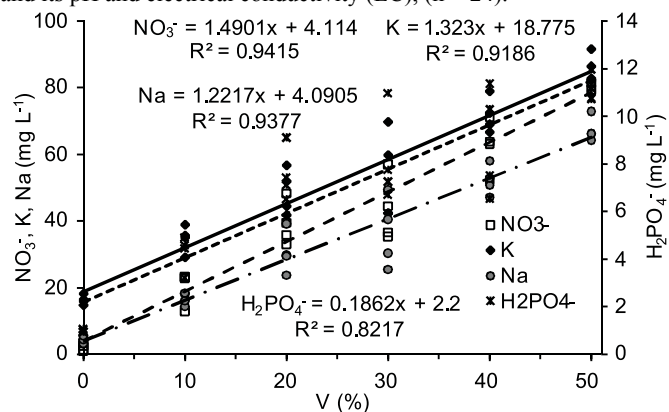


Fig. 3. Relationships between vermicompost (V) content in the substrate mixture and nitrate (NO_3^- , dashed line), potassium (K , solid line), sodium (Na , dashed-dotted line) and phosphorus ($H_2PO_4^-$, dotted line).

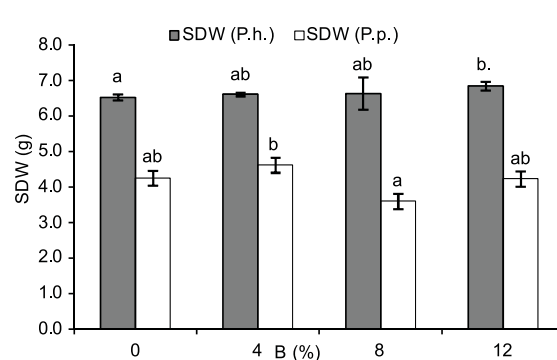
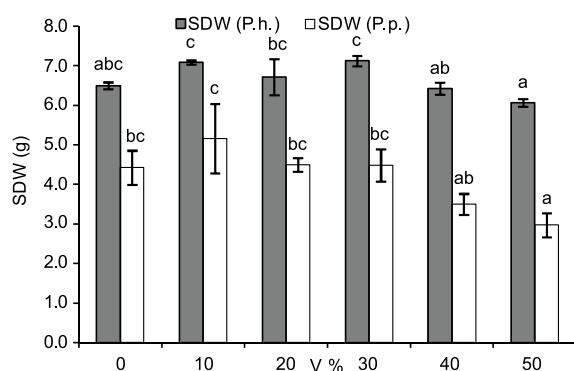


Fig. 4. Mean values (\pm SE) of shoot dry weight (SDW) for *Petunia* ($P.h.$) and *Pelargonium* ($P.p.$) grown in different doses of vermicompost (V) and biochar (B) in the substrate. Significance level: $p = 0.017$ for *Petunia* and $p = 0.044$ for *Pelargonium*. Different letters indicate significant differences between V rates for every species.

was added to the mixtures (Fig. 2, and A.5 in the appendix). EC and pH were positively related to V ratio ($P < 0.01$, $n = 24$) (Fig. 2). However, pH and EC were not significantly related to B .

Concentration of $N-NH_4^+$ tended to decrease with higher doses of B for all levels of V , and concentration of $N-NO_3^-$ increased ($r = 0.97$, $P < 0.001$, $n = 24$) with increasing rates of V (Fig. 3, and A.5 in the appendix).

Mixtures containing higher proportion of B and V had a higher organic matter content. Concentration of OM in all mixtures differed significantly from that in the control regardless of the amount of V and B in the mixture (Table 3).

Plant growth and flower production: Table 4 shows the biomass accumulated by the plants and the number of flowers per plant, for the two ornamental crops.

In general, B rates of 4 - 12 % with moderate V proportions 10 - 30 % tended to produce the highest SDW for *Petunia*, but 50 % V resulted in a slight negative effect. The overall trend for *Pelargonium* indicated that 40 - 50 % V mixture did not favor the growth and flowering (Fig. 4). Chlorosis symptoms were observed only in *Petunia* in the case of the mixture 38:50:12 and they were not very marked. Chlorosis was not observed in *Pelargonium*.

For *Petunia*, it can be noted that leaf concentrations of Ca , K , Mg and Na were directly related to their availability in the substrate ($r = 0.73$, 0.89 , 0.53 and 0.91 respectively, $P < 0.01$, $n = 24$). In *Pelargonium*, the leaf concentrations of Ca , K and Na were directly related to their availability in the substrate ($r = 0.53$, 0.50 and 0.95 respectively, $0.01 < P < 0.05$, $n = 24$), but an inverse correlation was observed between leaf Na concentration and SDW ($r = -0.58$, $P < 0.01$, $n = 24$). Additionally, in *Pelargonium*, inverse relationships were observed between SDW and available nutrient concentrations in the growth media: Ca ($r = -0.57$, $P < 0.01$, $n = 24$), K ($r = -0.63$, $P < 0.01$, $n = 24$), Mg ($r = -0.55$, $P < 0.01$, $n = 24$), Na ($r = -0.64$, $P < 0.01$, $n = 24$), $N-NO_3^-$ ($r = -0.63$, $P < 0.01$, $n = 24$) and P ($r = -0.54$, $P < 0.01$, $n = 24$).

Discussion

Substrate characteristics: Substrates used in production of horticultural crops in containers are predominantly constituted by organic components and their physical properties are key factors to identify strategies that can be implemented to reduce negative effects on crop growth (Bilderback *et al.*, 2005). We found in this work that there was a trend to a slight decrease

Table 4. Plant-growth parameters of *Petunia* and *Pelargonium* grown on different substrate mixtures (treatments).

Treatments	<i>Petunia</i>			<i>Pelargonium</i>		
	SDW	Flowers	Chlorosis	SDW	Flowers	Spots
S:V:B ^x	(g)	(n° flowers)	(range)	(g)	(flowers+buds) ^y	(range)
100:00:00	6.46 abcd	10.6 a	1.0	3.84 bcdef	0.77 bc	1.0
96:00:04	6.62 abcd	10.6 a	1.0	5.02 efg	0.91 bcd	1.2
92:00:08	6.64 abcd	11.8 ab	1.2	3.53 abcde	0.86 bcd	1.2
88:00:12	6.28 abcd	8.4 a	1.0	5.30 efg	0.97 bcd	1.6
90:10:00	6.94 abcde	9.6 a	1.0	5.20 fg	0.87 bcd	1.0
86:10:04	7.18 bcde	9.6 a	1.0	7.54 h	0.84 bc	1.2
82:10:08	7.12 cde	10.8 a	1.0	3.34 abcd	0.91 bcd	1.2
78:10:12	7.10 bcde	12.4 ab	1.0	4.56 bcdefg	1.06 cd	1.0
80:20:00	6.08 ab	9.0 a	1.0	4.54 defg	0.84 bc	1.2
76:20:04	6.14 abcd	9.2 a	1.0	4.64 defg	0.74 b	1.2
72:20:08	6.62 abcd	10.2 a	1.0	4.30 bcdefg	0.85 bc	1.0
68:20:12	8.04 e	17.0 b	1.0	4.50 cdefg	1.14 d	1.6
70:30:00	6.86 abcd	10.4 a	1.0	4.58 defg	1.01 bcd	1.2
66:30:04	7.4 de	8.4 a	1.0	5.60 fg	0.95 bcd	1.2
62:30:08	6.96 abcde	11.4 a	1.0	4.02 bcdef	0.83 bc	1.2
58:30:12	7.28 cde	13.0 ab	1.0	3.74 bcdef	0.90 bcd	1.6
60:40:00	6.82 abcd	10.8 a	1.0	3.80 bcdefg	0.90 bcd	1.0
56:40:04	6.178 abc	7.8 a	1.0	2.78 ab	0.35 a	1.0
52:40:08	6.18 abc	9.4 a	1.0	3.44 abcde	0.90 bcd	1.2
48:40:12	6.52 abcd	9.2 a	1.0	3.98 bcdefg	0.88 bcd	1.6
50:50:00	6.00 ab	8.8 a	1.0	3.54 abcde	0.73 b	1.6
46:50:04	6.14 abc	8.2 a	1.0	2.12 a	0.32 a	1.0
42:50:08	6.28 abcd	9.2 a	1.0	2.94 abc	0.78 bc	1.2
38:50:12	5.84 a	10.0 a	2.2	3.28abcd	0.82 bc	1.0
<i>p</i>	***	***	n.s	***	***	n.s

SDW: shoot dry weight.

^x S:V:B, Volume fraction of peat-based substrate (S), vermicompost (V) and biochar (B). Control = 100:00:00^y Transformed variable log 10*p*, significance level: *** indicates $p \leq 0.001$. Different letters in numerical columns indicate significant differences between treatments (T3-Dunnnett test). n.s.: not significant.

in *As* and an increase in *Db* with increasing *V* and *B* fractions. Being *V* and *B* more lightweight than *S*, it can be speculated that the substrates particles were filling the air gaps of the peat-based substrate. This resulted in a slightly less ideal substrate (Arancon and Edwards, 2005). However, considering mixtures containing $V \leq 30\%$, all of them were within the optimum range for *As*, while the deviation in *Db* was not very important in absolute value (Fonteno and Bilderback, 1993; Yeager *et al.*, 2000), taking into account that they were within the range of other nursery substrates like *Sphagnum* peat moss (0.06 to 0.12 kg dm⁻³), other peat mosses (0.08 to 0.28 kg dm⁻³), conifer barks (0.20 to 0.40 kg dm⁻³), coconut fibers (0.18 to 0.20 kg dm⁻³) or vermiculite (0.06 to 0.17 kg dm⁻³) (Landis, 1990; Harp, 2011).

pH of the control substrate was slightly increased by *V*. These changes in pH coincided with those reported by Tyler *et al.* (1993) according to which the pH increased in response to increasing concentrations of composted turkey litter added to a plant container medium. Ideal pH levels range for *Petunia* are from 5.5 to 6.2, and from 6.2 to 6.8 for *Pelargonium* (Irwin, 2002). With the exception of mixture 52:40:08 (with pH = 6.1), other mixtures (containing $V \geq 40\%$, or $B = 12\%$ together with $V = 20\%$ or 30%) had pH values higher than 6.2, but chlorosis symptoms were not observed (except for 38:50:12 mixture with *Petunia*).

Although pH was below 6.2 for some mixtures, chlorosis was not observed in *Pelargonium*. Therefore, based on the *Petunia* pH range, less than 40 % *V* should be used. Mixtures with $V \leq 10\%$ might take a higher dose of *B* without exceeding the recommended pH limits for growing *Petunia*. Mixtures without *V* (i.e. $V = 0\%$) might take a higher dose of *B* without exceeding the recommended pH limits for growing *Pelargonium* and *Petunia*. The positive relationship between EC and *V* can be explained by the high EC of vermicompost. Similar results were reported by Atiyeh *et al.* (2001). Klock (1997) reported an increase in EC of 1.3 to 2.8 times over the control treatment with the addition of vermicompost. In the present study, EC increased 5.7 times over the peat-based substrate in mix 38:50:12.

Organic matter from the control was 55.3 % and gradually increased up to 73.7 % in substrate 38:50:12 because of the addition of vermicompost and biochar to the mixtures. In substrates containing $V \leq 10\%$, OM concentration was slightly more influenced by *B* content than by the *V* content.

Plant growth: For both species, SDW decreased for $V \geq 40\%$, but to a greater extent for *Pelargonium* than for *Petunia*. This could be due to several reasons, such as increased EC and the decrease in *As*. *Pelargonium* was more affected by its higher sensibility to substrate salinity. Mixes with lower *As* (Milks *et*

al., 1989) and higher pH and EC levels tended to induce lower SDW. Similar results were reported by Sultana *et al.* (2015) who observed that shoot height and total number of flowers of *Zinnia elegans* increased when grown in mixtures containing (10–20) % of vermicompost. On the other hand, in our work, *B* caused a lesser effect than *V* on substrate properties and on plant growth and nutrition, probably due to the lower amounts of *B* applied.

Overall, nutrient concentrations in the leaves were within the usual ranges suggested for these species (Mills and Jones, 1999), and did not show clear deficiency symptoms. The high Na leaf concentration in *Petunia* gives us an indication of its high salt tolerance, and the low Na leaf concentration in *Pelargonium* is typical of not tolerant species, because Na is not an essential nutrient for these plants and may be toxic (Hund-Rinke, 2008). The decrease in N, Fe and Mn for *Pelargonium* as *V* increased is characteristic when toxic levels of nutrients are present in growth media (Marschner, 1998), probably due to the effect of growth media salinity due to the dissolved mineral ions.

Environmental effect: Some studies have shown reductions in GHG emissions when *B* (Steiner and Harttung, 2014) is used as peat substitute for growing plants. *B* decomposes slowly (Kuzakov *et al.*, 2009) and can be stored for relatively long periods. *V* has a faster decomposition rate, so no significant C sequestration or storage in soil is expected by *V*, and this is why we only are going to calculate GHG emissions based in the biochar potential effect. Nevertheless, as peat volume substituted by *V* has a CO₂ sink role and, in addition, *V* contains mineral nutrients that potentially reduce the use of inorganic fertilizers contributing to reduce CO₂ emissions and energy consumption (Audsley *et al.*, 2003), *V* has been included in our calculation. Thus, this study is focused on the biochar effect to calculate how gaseous emissions associated with peat decomposition can at least be avoided if peat is substituted by *B*. The data presented herein shows that it is possible to grow *Petunia* and *Pelargonium* by replacing a portion of peat in a peat based substrate with a mixture of *V* and *B* at ranges up to 30 % *V* and 12 % *B*. It would be possible to save up to 117.8 kg of peat per tonne of substrate by substituting it with *V* and *B* taking into account bulk density of those materials (135, 206.9 and 131) kg m⁻³ for *P*, *B* and *V* respectively, and their weight to weight ratio in the mixture (47.7 %, 15.1 %, and 24.0 %, respectively). Thus, up to 151.4 kg of biochar and 239.6 kg of vermicompost may substitute 117.8 kg of peat in the new mixed substrate. The replacement of peat-moss with biochar could avoid up to 3.25 t of carbon dioxide equivalent (CO_{2e}) per tonne of peat substituted (Steiner and Harttung, 2014). Under the above mentioned assumption, the use of biochar could save up to 624.2 kg of CO_{2e} per tonne of the new substrate. Considering the mix 58:30:12 (*S*:*V*:*B*, volume basis) and its obtained *Db* measurement, it will be possible to store up to 88.74 gr of CO_{2e} per 800 ml container for long periods of time, first in the plant's growing container and then in the soil after transplanting (no C storage has been calculated when transplanting seedlings to containers because in seedling trays no peat substitution by vermicompost and biochar happened).

As shown in the present work, *V* and *B* can be mixed together in a substrate (hypothesis *a*). Both are renewable resources. *V* provides fertility and reduces inorganic mineral fertilization, and *B* contributes to carbon fixation in the long term. We have also

partially verified hypothesis *b*), that an optimal range of *B* and *V* ratios will be obtained to grow these species. The top *V* rates (40 % to 50 %) should not be reached as it was reported by García-Albarado *et al.* (2010) and Sardoei, (2104). Nevertheless more research will be needed to verify how these species will grow with (0–30) % *V* mixes and higher ratios of *B* than 12%. Finally, it is possible to state that hypothesis *c*) has been proven as a number of treatments produced plants of the same or better commercial quality than plants grown in the control peat-based treatment.

The data presented support the following conclusions: It is possible to grow containerized *Petunia hybrida* and *Pelargonium peltatum* plants with commercial quality after 3 or 4 months of cultivation, using substrates comprising a peat-based substrate mixed with biochar and/or vermicompost. As much as 30 % by volume of *V* and 12 % of *B* could be used in the substrate mixture without any adverse effects on plant growth and flower production. However, one must avoid adding the maximum doses of *V* (40 to 50) % for growing *Pelargonium* and 50 % *V* for *Petunia*. Biochar and vermicompost offer great environmental advantages in their use as a peat-based growing media replacement in ornamental plant production because their C storage and / or CO₂ emission reduction.

The use of biochar and vermicompost is also compatible with the maintenance of the ornamental quality required for cultivated plants. Nevertheless more research would be necessary to a wider range of crops and with more standardized biochar and vermicompost products.

Acknowledgments

This work was partially supported by the project CTQ 2013-46804-C2-1-R of the Spanish Ministry of Economy and Competitiveness and the European Regional Development Funds (ERDF). The author wish to thank the Horticultural Department and Carbon Sequestration and Management Center of Ohio State University for providing materials and facilities for this investigation, also he is deeply grateful to Mrs. Loewe and Dr. J. Altland from Application Technology Research Unit at Wooster OSU campus for their laboratory assistance in determining substrates mixtures physical properties.

References

- Alexander, P., N. Bragg, R. Meade, G. Padelopoulos and O. Watts, 2008. Peat in horticulture and conservation: the UK response to a changing world. *Mires and Peat*, 3: 1-11.
- Ansorena Miner, J., 1994. *Sustratos propiedades y caracterización*. Mundi-Prensa. Madrid (Spain). 141 p.
- Arancon, N. Q., Edwards, C. a., Bierman, P., Metzger, J. D. and C. Lucht, 2005. Effects of vermicomposts produced from cattle manure, food waste and paper waste on the growth and yield of peppers in the field. *Pedobiologia*, 49(4), 297–306. <https://doi.org/10.1016/j.pedobi.2005.02.001>
- Atiyeh, R., N. Arancon, C. Edwards and J. Metzger, 2000. Influence of earthworm-processed pig manure on the growth and yield of greenhouse tomatoes. *Bioresource Technology*, 75: 175-180. doi:10.1016/S0960-8524(00)00064-X.
- Audsley, E. (coord.), 2003. Harmonisation of environmental life cycle assessment for agriculture, Final Report. Concerted Action AIR3-CT94-2028, European Commission, Directorate-General VI Agriculture, Brussels, B-1049 Belgium, 129 p.

- Beck, D.A., G.R. Johnson and G.A. Spolek, 2011. Amending greenroof soil with biochar to affect runoff water quantity and quality. *Environmental pollution*, 159: 8-9, 2111-8.
- Bilderback, T.E., S.L. Warren, J.S. Owen and J. P. Albano. 2005. Healthy Substrates Need Physicals Too. *HortTechnology*, 15: 747-751.
- Dahlquist, R.L. and J.W. Knoll. 1978. Inductively Coupled Plasma-Atomic Emission Spectrometry: Analysis of biological materials and soils for major trace and ultra-trace elements. *Appl. Spectroscopy*, 32: 1-30.
- Dispenza, V.; De Pasquale, C.; Fascella, G.; Mammano, M. M.; Alonzo, G. 2016. Use of biochar as peat substitute for growing substrates of Euphorbia × lomi potted plants. *Spanish Journal of Agricultural Research*, Volume 14, Issue 4, e0908. <http://dx.doi.org/10.5424/sjar/2016144-9082>.
- Do, T. C. V. and H. W. Scherer. 2013. Compost as growing media component for salt-sensitive plants, *Plant Soil Environ*, 59: 214-220.
- DOI-USGS (U.S. Department of the Interior - U.S. Geological Survey). 2013. <https://minerals.usgs.gov/minerals/pubs/commodity/peat/>
- Fisher, D. and B. Glaser. 2012. Synergisms between compost and biochar for sustainable soil amelioration. In *Management of organic waste*. InTech. doi: 10.5772/31200.
- Fonteno, W.C. and T.E. Bilderback. 1993. Impact management of hydrogel on physical properties of coarse-structured horticultural substrates. *J. Amer. Soc. Hort. Sci.*, 118(2): 217-222.
- García-Albarado, J.C., L.I. Trejo-Téllez, M.A. Velásquez-Hernández, A. Ruiz-Bello and F.C. Gómez-Merino, 2010. Crecimiento de petunia en respuesta a diferentes proporciones de composta en sustrato. *Rev. Chapingo. Ser. Hortic.*, 16: 107-113. ISSN: 1027-152X.
- Graber, E.R., Y.M. Harel, M. Kolton, E. Cytryn, A. Silber, D.R. David, L. Tsechansky, M. Borenshtein and Y. Elad. 2010. Biochar impact on development and productivity of pepper and tomato grown in fertirigated soilless media. *Plant Soil*, 337: 481-496.
- Gu, M., Li, Q., Steele, P.H., Niu, G., Yu, F., 2013. Growth of "Fireworks" gomphrena grown in substrates amended with biochar. *J. Food, Agric. Environ.* 11, 819-821.
- Harp, D., 2011. Container grown plant production, in: Gawel, N. (Ed.), *Proceedings of the Southern Nursery Association, Research Conference*, Vol. 56, 235-302.
- Houba, V.J.G., J. Uittenbogaard and P. Pellen. 1996. Wageningen evaluating programmes for Analytical Laboratories (WEPAL), organisation and purpose. *Commun. Soil Sci. Plant Anal.*, 27: 421-431.
- Hund-Rinke, K.. 2008. Benefit of ecotoxicological tests for the characterization of composts. CODIS International congress. 137-144.
- Irwin, J., 2002. Optimal pH requirements for different species. *Minnesota Commercial Flower Growers Bul.* April pg11. Dep. of Horticultural Science, University of Minnesota. <http://www.florifacts.umn.edu/mnfgbulletins/April%202002%20Bulletin/pH%20requirements.pdf>
- Jeffery, S., Verheijen, F.G.A., van der Velde, M., Bastos, A.C., 2011. A quantitative review of the effects of biochar application to soils on crop productivity using meta-analysis. *Agric. Ecosyst. Environ.*, 144(1): 175-187. doi:10.1016/j.agee.2011.08.015.
- Klock, K.A., 1997. Growth of salt sensitive bedding plants in media amended with composted urban waste. *Compost Sci. Util*, 5: 55-59. doi: 10.1080/1065657X.1997.10701886
- Kuzyakov, Y., I. Subbotina, H. Chen, I. Bogomolova and X. Xu, 2009. Black carbon decomposition and incorporation into soil microbial biomass estimated by ¹⁴C labeling. *Soil Biol. Biochem.*, 41: 210-219, doi:10.1016/j.soilbio.2008.10.016.
- Laird, D. A., 2008. The Charcoal Vision: A win-win-win scenario for simultaneously producing bioenergy, permanently sequestering carbon, while improving soil and water quality. *Agronomy Journal*, 100(1): 178-181. doi:10.2134/agronjnl2007.0161.
- Landis, T.D., 1990. Containers and growing media, Vol. 2, *The container tree nursery manual*. Agric. Handbook. 674. Washington, DC. U. S. Department of Agriculture, Forest Service. 41-85.
- Lal, R., 2011. Sequestering carbon in soils of agro-ecosystems. *Food Policy* 36: S33-S39. doi:10.1016/j.foodpol.2010.12.001
- Lal, R., 2016. Biochar and soil carbon sequestration, in: Guo, M., He, Z., Uchimiya, S.M. (Eds.), *Agricultural and environmental applications of biochar: advances and barriers*. SSSA Special Publication 63, Madison, WI:175-197. doi: 10.2136/sssaspecpub63.2014.0042.5
- Lehmann, J., 2007. Bio-energy in the black. *Frontiers in Ecology and the Environment* 5(7): 381-387. doi:10.1890/060133
- López R, F. Cabrera, E. Madejón, F. Sancho and J.M. Álvarez, 2008. Urban Composts as an Alternative for Peat in Forestry Nursery Growing Media. In: Hao X, editor. *Compost I. Dynamic Soil, Dynamic Plant*, 1: 60-66.
- Marschner, H., 1998. *Mineral nutrition of higher plants*. 2nd. Ed., Academic Press, London, UK, 889 p. ISBN: 0-12-473543-6
- Michel, J., 2010. The physical properties of peat: a key factor for modern growing media. *Mires and Peat* 6: 2-7.
- Milks, R.R., W.C. Fonteno and R.A. Larson, 1989. Hydrology of horticultural substrates: III. Predicting air and water content of limited volume plug cells. *J. Amer. Soc. Hort. Sci.*, 114: 57-61.
- Miller, R.O., 1998. Microwave digestion of plant tissue in a closed vessel, in: Kalra, Y.P. (Ed.), *Handbook and reference methods for plant analysis*. CRC Press, Boca Raton New York.
- Mills, H.A. and J.B. Jones. 1996. *Plant Analysis Handbook II. A Practical Sampling, Preparation, Analysis and Interpretation Guide*, MicroMacro Publishing, Inc.: Athens, Georgia.
- Mionk, R.W. and H.H. Wiebe, 1961. Salt tolerance & protoplasmic salt hardness of various woody & herbaceous ornamental plants. *Plant Physiology*, 36(4): 478.
- Mukherje, A. and R. Lal, 2014. The biochar dilemma. *Soil Research*, 52: 217-230. doi: 10.1071/SRI3359
- Perry, A., 2011. Carefully unraveling the intricacies of biochar. *Agricultural Research magazine*, 59(10): 4-8. <http://agresearchmag.ars.usda.gov/2011/nov/biochar>
- Sardoei, A., 2014. Vermicompost effects on the growth and flowering of marigold (*Calendula officinalis*). *European Journal of Experimental Biology*, 4(1): 651-655.
- Schmidt, H.P., C. Kammann, C. Niggli, M.W.H. Evangelou, K.A. Mackie and S. Abiven, 2014. Biochar and biochar-compost as soil amendments to a vineyard soil: Influences on plant growth, nutrient uptake, plant health and grape quality. *Agriculture, Ecosystems and Environment*, 191: 117-123. doi:10.1016/j.agee.2014.04.001.
- Sohi, S., J.L. Gaunt and J. Atwood, 2013. Biochar in growing media: A sustainability and feasibility assessment. *UK Biochar Research Center, University of Edinburgh*, 84 p.
- Steiner, C. and T. Hartung, 2014. Biochar as growing media additive and peat substitute. *Solid Earth Discussions* 6(1): 1023-1035. doi:10.5194/sed-6-1023-2014
- Sultana, S., A. Kashem, A. Kalam and M. Moniruzzaman, 2015. Comparative assessment of cow manure vermicompost and NPK fertilizers and on the growth and production of zinnia (*Zinnia elegans*) flower. *Open Journal of Soil Science*, 5: 193-198. doi:10.4236/ojss.2015.59019
- Tian, Y., X. Sun, S. Li, H. Wang, L. Wang, J. Cao and L. Zhang. 2012. Biochar made from green waste as peat substitute in growth media for *Calathea rotundifolia* cv. Fasciata. *Sci. Hortic.*, 143: 15-18. doi:10.1016/j.scienta.2012.05.018.
- Tyler, H. H., Warren, S. L., Bilderback, T. E., & Fonteno, W. C. 1993. Composted turkey litter: I. Effect on chemical and physical properties of a pine bark substrate. *Journal of Environmental Horticulture*, 11, 131-131. Retrieved from http://hrresearch.org/docs/publications/JEH/JEH_1993/JEH_1993_11_3/JEH_11-3-131-136.pdf
- Yeager, T., C. Gilliam, T.E. Bilderback, D. Fare, A. Niemiera and K. Tilt, 2000. Best management practices guide for producing container-grown plants. *Southern Nurserymen's Association*, Atlanta, USA, 69 p.

APPENDIX

Table A.1. Biochar (*B*) characterization (Soil Reef Pure 02 by Soil Control Lab). International Biochar Initiative (IBI) Level I

Component	Dry basis	unit	Method	Particle Size Distribution ASTM D2862 granular	Retained (%)	Fraction (%)
Total Ash	8.4	%	ASTM D1762-84 (750c)	(mm)		
Organic Carbon	75.8	%	CHN by dry combustion	> 19	0.0	0.0
Inorganic Carbon	0.45	%	HCl treated	16-19	0.0	0.0
Hydrogen/Carbon (H:C)	0.48	molar ratio		9.5-16	0.0	0.0
Hydrogen	3.0	%	CHN by dry combustion	6.3-9.5	0.0	0.0
Total Nitrogen	0.45	%	CHN by dry combustion	4.0-6.3	0.4	0.4
Total Oxygen	20.2	%	by difference	2.0-4.0	23.0	22.5
Liming (neut.value)	4.7	%CO ₃ Ca	Rayment & Higginson	1.0-2.0	53.9	31.0
Liming (carbonate.value)	3.8	%CO ₃ Ca	ASTM D4373	0.425-1.0	86.8	32.9
Activity (Butane)	7.6	g/100g	ASTM D5742 (butane)	< 0.425	100	13.2
Bulk density	206.9	kg m ⁻³				
Sulfur	0.094	%				
Energy (HHV)	27791	kJ/kg				
Moisture	12.7	%	ASTM D1762-84 (105c)			

Table A.2. Element content in biochar (*B*) (Soil Reef Pure 02 by Soil Control Lab). International Biochar Initiative (IBI) Level II

Component	dry basis	Unit	Method
Arsenic (As)	9.8	mg kg ⁻¹	Bureau de Normalisation de Quebec
Cadmium (Cd)	0.17	mg kg ⁻¹	Amlinger, Faroino and Pollack (2004)
Chromium (Cr)	28	mg kg ⁻¹	Amlinger, Faroino and Pollack (2004)
Cobalt (Co)	4.6	mg kg ⁻¹	Bureau de Normalisation de Quebec
Copper (Cu)	23	mg kg ⁻¹	Amlinger, Faroino and Pollack (2004)
Lead (Pb)	12	mg kg ⁻¹	Amlinger, Faroino and Pollack (2004)
Molybdenur (Mo)	< 0.2	mg kg ⁻¹	Bureau de Normalisation de Quebec
Mercury (Hg)	< 0.2	mg kg ⁻¹	Amlinger, Faroino and Pollack (2004)
Nickel (Ni)	17	mg kg ⁻¹	Amlinger, Faroino and Pollack (2004)
Selenium (Se)	< 0.2	mg kg ⁻¹	Bureau de Normalisation de Quebec
Zinc (Zn)	82	mg kg ⁻¹	Amlinger, Faroino and Pollack (2004)
Boron (Bo)	117	mg kg ⁻¹	Test Meth. Exam. Compost and Composting (2001)
Chlorine (Cl)	1154	mg kg ⁻¹	Test Meth. Exam. Compost and Composting (2001)
Sodium (Na)	5194	mg kg ⁻¹	Test Meth. Exam. Compost and Composting (2001)
Potassium (K) Total	20	%	Enders and Lehmann (2004)
Phosphorus (P) Total	0.37	%	Enders and Lehmann (2004)
Ammonia (NH ₄ -N)	5.7	mg kg ⁻¹	Rayment & Higginson
Nitrate (NO ₃ -N)	64	mg kg ⁻¹	Rayment & Higginson
Moisture	12.7	%	Test Meth. Exam. Compost and Composting (2001)

Table A.3. Vermicompost (*V*) characterization label information

Component	Dry basis	units	Component	Dry wt.	units
Total Nitrogen:	2.9	%	Lime as CaCO ₃	4450	mg kg ⁻¹
Ammonia (NH ₄ -N):	17	mg kg ⁻¹	Organic Matter:	72.7	%
Nitrate (NO ₃ -N):	3100	mg/kg	Organic Carbon:	35.0	%
Org. Nitrogen (Org.-N):	2.6	%	Ash:	27.3	%
Phosphorus (as P ₂ O ₅):	1.0	%	C/N Ratio	12	ratio
Potassium (as K ₂ O):	0.65	%	AgIndex	10	ratio
Calcium (Ca):	2.4	%	Copper (Cu):	170	mg kg ⁻¹
Magnesium (Mg):	0.88	%	Iron (Fe):	5500	mg kg ⁻¹
Sulfate (SO ₄ -S):	520	mg kg ⁻¹	Lead (Pb):	2.3	mg kg ⁻¹
Boron (Total B):	49	mg kg ⁻¹	Manganese (Mn):	250	mg kg ⁻¹
Moisture:	0	%	Mercury (Hg):	< 1.0	mg kg ⁻¹
Sodium (Na):	0.30	%	Molybdenum (Mo):	4.2	mg kg ⁻¹
Chloride (Cl):	0.16	%	Nickel (Ni):	27	mg kg ⁻¹
pH Value:	NA	unit	Selenium (Se):	1.2	mg kg ⁻¹
Bulk Density :	131.0	kg m ⁻³	Zinc (Zn):	910	mg kg ⁻¹

Table A.4. Standard peat based growing media (S) label information (mg kg⁻¹, except for pH)

Component	Dry basis	Component	Dry basis
pH	5.5-6.5	B	0.0-0.15
NH ₄ -N	0.0-50	Cu	0.0-0.12
NO ₃ -N	50-150	Fe	0.5-5.0
P	5.0-40	Mn	0.0-4.0
K	100-300	Mo	0.0-0.15
Ca	50-200	Na	20-50
Mg	40-200	S	100-250
Zn	0.0-1.0		

Table A 5. Selected physico-chemical properties of different substrate mixtures (treatments). Units: mg L⁻¹ for nutrients and mS m⁻¹ for EC

Treatment	pH	EC	N-NH ₄	NN-NO ₃	H ₂ PO ₄	K	Ca	Mg	SO ₄ ⁻²	Na	Fe
S:V:B ¹		(mS m ⁻¹)	(mg L ⁻¹)								
100:00:00	5.47	14.2	3.06	3.6	1.07	15.89	5.15	4.84	7.35	3.23	0.02
96:00:04	5.35	13.1	1.30	2.5	0.95	16.50	4.61	4.34	7.06	3.39	0.03
92:00:08	5.60	10.8	0.91	1.3	0.96	14.88	4.05	2.61	5.86	4.55	0.01
88:00:12	5.63	10.8	0.15	1.0	1.09	18.20	7.54	3.35	7.61	5.68	<0.01
90:10:00	5.46	31.7	0.18	23.0	4.47	29.29	13.13	11.60	7.13	14.54	<0.01
86:10:04	5.81	35.9	0.18	23.2	4.93	35.04	13.11	10.83	8.81	18.29	0.03
82:10:08	5.84	334	0.22	22.9	4.60	35.69	13.33	11.75	10.06	16.24	0.02
78:10:12	5.93	27.4	0.16	13.0	3.23	38.95	6.41	3.87	6.97	18.52	<0.01
80:20:00	5.76	47.1	0.16	39.6	7.45	44.59	19.74	15.33	8.04	29.74	0.01
76:20:04	5.82	32.4	0.13	48.4	9.10	56.76	23.30	18.28	9.90	39.13	0.02
72:20:08	5.66	40.2	0.24	33.0	7.12	41.80	16.66	13.77	8.51	23.88	0.01
68:20:12	6.22	49.8	0.17	35.9	6.53	51.98	15.14	12.66	8.96	29.60	0.01
70:30:00	5.87	46.8	0.19	36.7	7.24	42.48	18.56	15.67	8.26	25.57	0.02
66:30:04	6.06	50.0	0.41	35.3	6.72	42.45	21.98	16.46	10.97	30.25	0.02
62:30:08	6.11	51.6	0.14	56.9	10.98	69.81	28.06	21.44	13.19	49.55	0.01
58:30:12	6.27	51.1	0.06	44.6	7.77	59.81	21.96	14.31	9.72	40.49	0.01
60:40:00	6.42	76.0	0.22	63.0	6.52	72.53	24.25	17.10	10.74	50.91	0.02
56:40:04	6.22	54.6	0.32	63.9	10.32	69.02	26.75	20.35	10.54	53.03	0.02
52:40:08	6.07	64.9	0.14	71.1	11.34	78.99	29.55	23.00	12.69	58.15	0.01
48:40:12	6.38	62.3	0.09	52.4	7.49	66.68	20.37	15.41	8.15	47.29	0.01
50:50:00	6.28	88.6	0.25	81.5	11.98	82.79	36.31	26.61	11.83	66.14	0.03
46:50:04	6.26	79.0	0.23	79.1	10.76	82.22	31.99	24.28	11.90	64.16	0.02
42:50:08	6.31	78.5	0.09	79.3	10.70	86.41	31.81	23.55	12.22	66.06	0.01
38:50:12	6.57	73.3	0.06	81.5	11.2 0	91.66	36.60	25.83	16.81	72.78	0.01

¹ S:V:B, Volume fraction of peat based substrate (S), vermicompost (V) and biochar (B). Control: 100:00:00.Table A.6. Chlorosis level and spots ranges visually estimated in *Petunia* and *Pelargonium* leaves

Code	Chlorosis level	Spots
1	No chlorosis green plant	No spots
2	Light chlorosis on terminal leaves	1-9 % leaf's surface covered by spots
3	Medium chlorosis on terminal leaves	10-19% leaf's surface covered by spots
4	Intense chlorosis on terminal leaves	20-29% leaf's surface covered by spots
5	Light chlorosis on terminal leaves+ remaining leaves	30-39% leaf's surface covered by spots
6	Medium chlorosis on terminal leaves+ remaining leaves	40-49% leaf's surface covered by spots
7	Intense chlorosis on terminal leaves+ remaining leaves	50-59% leaf's surface covered by spots
8	Very intense chlorosis on terminal leaves	60-69% leaf's surface covered by spots
9	Very intense chlorosis on terminal leaves+ remaining leaves	70-79% leaf's surface covered by spots
10	Yellowish plant	80-100% leaf's surface covered by spots

Table A-7. Leaf mineral concentrations (dry weight basis) of *Petunia* grown on different biochar and vermicompost mixtures

Treatment	N	P	K	Ca	Mg	S	Fe	Mn	B	Cu	Zn	Na
<i>S:V:B</i> ¹			(%)						($\mu\text{g g}^{-1}$)			
100:00:00	4.55	0.64	2.66	0.93	0.56	0.76	192.3	104.2	33.8	12.3	64.4	293
96:00:04	4.28	0.59	2.96	0.86	0.55	0.76	167.3	110.6	29.6	10.9	61.7	324
92:00:08	4.05	0.56	3.23	0.85	0.55	0.83	151.5	114.3	28.2	10.2	65.5	372
88:00:12	3.79	0.58	3.52	1.05	0.63	0.93	175.7	120.6	31.1	8.7	71.3	373
90:10:00	4.21	0.76	3.36	1.26	0.71	0.69	189.4	37.5	33.3	13.0	83.9	689
86:10:04	3.90	0.73	3.83	1.14	0.72	0.64	118.8	43.1	29.1	14.8	81.9	725
82:10:08	4.20	0.73	3.43	1.29	0.73	0.74	210.6	47.9	30.1	12.5	86.5	674
78:10:12	3.96	0.54	3.85	1.24	0.62	0.76	187.0	77.1	28.3	14.9	68.0	726
80:20:00	4.40	0.89	3.99	1.38	0.67	0.66	153.8	44.3	33.4	15.8	99.1	889
76:20:04	3.98	0.77	3.87	1.24	0.66	0.63	166.3	43.4	34.2	9.9	91.9	788
72:20:08	4.31	0.77	3.47	1.43	0.69	0.65	211.1	56.1	38.0	12.5	83.5	804
68:20:12	3.91	0.70	3.51	1.26	0.62	0.67	164.2	57.4	33.5	13.1	83.6	777
70:30:00	4.20	0.77	3.61	1.26	0.67	0.65	189.0	36.8	33.8	15.1	88.6	783
66:30:04	4.28	0.83	4.11	1.33	0.68	0.66	150.1	49.0	34.3	17.1	99.0	940
62:30:08	4.07	0.76	4.13	1.41	0.71	0.69	158.0	57.2	39.9	10.5	104.5	1080
58:30:12	4.03	0.72	4.11	1.38	0.65	0.70	135.4	62.1	33.3	11.9	104.0	1027
60:40:00	3.91	0.77	4.28	1.27	0.62	0.62	173.1	46.0	39.2	11.5	110.5	968
56:40:04	4.15	0.82	4.25	1.41	0.66	0.71	142.9	59.0	37.6	13.3	117.7	1058
52:40:08	4.18	0.78	4.10	1.42	0.66	0.67	169.1	66.1	33.7	16.6	108.0	1016
48:40:12	4.23	0.75	4.20	1.52	0.70	0.72	192.7	80.2	36.1	7.3	119.3	1193
50:50:00	3.98	0.81	4.55	1.29	0.64	0.65	175.6	50.9	35.3	9.2	118.3	1070
46:50:04	4.25	0.80	4.33	1.43	0.67	0.71	203.9	71.7	39.9	11.6	123.6	1130
42:50:08	4.18	0.80	4.38	1.48	0.68	0.70	178.1	77.4	36.5	13.2	135.1	1148
38:50:12	4.09	0.70	4.50	1.30	0.67	0.70	181.9	65.0	42.7	10.6	96.3	1086
Average	4.13	0.73	3.84	1.27	0.66	0.70	172.4	65.7	34.4	12.4	94.4	831
(SE)	(0.04)	(0.02)	(0.10)	(0.04)	(0.01)	(0.01)	(4.7)	(5.0)	(0.8)	0.5	(4.2)	(56)
Sug.	3.85	0.47	3.13	1.20	0.36	0.33	84	44	18	3	33	3067
Range ²	7.60	0.93	6.68	2.81	1.37	0.80	168	177	43	19	85	10896

¹ *S:V:B*, Volume fraction of peat based substrate (*S*), vermicompost (*V*) and biochar (*B*). Control: 100:00:00. ² Suggested ranges (Mills and Jones, 1996).

Table A.8. Leaf mineral concentrations (dry weight basis) of *Pelargonium* grown on different biochar and vermicompost- based substrates

Treatment	N	P	K	Ca	Mg	S	Fe	Mn	B	Cu	Zn	Na
<i>S:V:B</i> ¹			(%)						($\mu\text{g g}^{-1}$)			
100:00:00	3.79	0.46	2.78	1.04	0.55	0.37	85.2	168.8	39.6	4.04	59.4	0.24
96:00:04	3.71	0.43	2.93	1.02	0.52	0.37	90.3	170.2	39.1	4.20	53.1	0.24
92:00:08	3.44	0.42	3.02	1.09	0.54	0.35	87.5	181.4	42.0	5.10	55.4	0.26
88:00:12	2.92	0.41	3.29	1.21	0.61	0.31	76.1	207.1	42.2	4.41	45.5	0.25
90:10:00	3.07	0.53	3.11	1.28	0.58	0.31	86.1	64.9	49.3	5.47	51.1	0.38
86:10:04	3.11	0.52	3.30	1.28	0.58	0.30	72.3	89.6	46.2	6.05	48.9	0.42
82:10:08	3.42	0.55	3.07	1.28	0.58	0.33	73.0	96.2	52.4	4.77	48.9	0.42
78:10:12	2.81	0.45	3.58	1.35	0.52	0.29	69.8	99.1	39.4	3.19	38.5	0.45
80:20:00	3.06	0.51	3.26	1.31	0.56	0.27	70.1	57.9	52.6	5.53	48.1	0.55
76:20:04	2.97	0.56	3.37	1.31	0.57	0.28	69.7	61.4	46.3	5.94	52.2	0.50
72:20:08	3.10	0.53	3.41	1.31	0.57	0.29	65.4	71.5	49.1	4.11	49.2	0.48
68:20:12	2.91	0.45	5.04	1.35	0.59	0.23	82.4	66.0	42.5	3.73	40.7	0.55
70:30:00	2.93	0.54	3.20	1.27	0.59	0.28	72.1	57.1	49.6	4.77	48.5	0.53
66:30:04	2.80	0.55	3.34	1.34	0.56	0.26	65.0	51.7	51.7	4.89	47.7	0.58
62:30:08	2.90	0.54	3.40	1.35	0.56	0.27	65.5	62.7	56.6	4.14	53.0	0.66
58:30:12	2.92	0.47	4.70	1.37	0.58	0.23	69.8	56.6	46.1	3.45	39.9	0.63
60:40:00	2.81	0.50	3.42	1.28	0.54	0.27	69.8	44.5	56.4	5.63	49.8	0.67
56:40:04	3.15	0.53	3.58	1.33	0.56	0.28	108.0	70.2	58.0	4.91	48.6	0.73
52:40:08	2.96	0.46	3.33	1.27	0.53	0.27	84.3	59.3	52.6	4.57	46.4	0.63
48:40:12	2.79	0.44	4.71	1.30	0.55	0.21	73.6	46.8	54.8	3.32	37.2	0.69
50:50:00	2.79	0.47	3.49	1.34	0.55	0.27	58.6	43.9	58.0	6.10	48.3	0.71
46:50:04	2.93	0.49	3.60	1.31	0.56	0.27	62.0	54.3	57.4	4.65	47.9	0.75
42:50:08	2.93	0.40	3.51	1.23	0.52	0.27	64.9	47.7	49.0	3.72	40.3	0.71
38:50:12	2.62	0.44	4.61	1.26	0.54	0.20	69.9	33.5	50.0	3.25	36.6	0.74
Average	3.04	0.49	3.54	1.27	0.56	0.28	74.6	81.8	49.2	4.58	47.3	0.53
(SE)	(0.06)	(0.01)	(0.12)	(0.02)	(0.01)	(0.01)	(2.3)	(10.0)	(1.3)	(0.18)	(1.2)	(0.03)
Sug.	3.3	0.30	2.50	0.80	0.20	0.25	100	40	30	5	7	--
Range ²	4.8	1.24	6.26	2.40	0.51	0.70	580	325	75	25	100	

¹ *S:V:B*, Volume fraction of peat based substrate (*S*), vermicompost (*V*) and biochar (*B*). Control: 100:00:00

² Suggested ranges (Mills and Jones, 1996).